Experiment 1F - Bubble Column Aeration

Responsible Authors:

Alex Nageli, Virginia Tech (amnageli@vt.edu)
Aidan Jackson, University of Washington (aidanj@uw.edu)
Group 35
July 15th, 2019

Abstract

The use of bacteria or other microorganisms in production processes is a growing trend across multiple industries. [5] In these cases, microorganisms offer the advantage of carrying out specific chemical reactions as part of their metabolism. These reactions are often more energy and material efficient than alternative production methods, in addition to producing less toxic waste. [5] The biochemical industry commonly takes advantage of unique or gene-edited strains to produce high purity and commodity chemicals at cheap cost. [1] Studies have predicted biopharmaceuticals, the majority of which are produced from bacteria-involved processes, to make up over 50% of all drugs in development in the next 5-10 years.^[4] The food industry has also been the most historic exploiter of bacteria-involved processes, with use in the production of yeast, yogurt, and alcohols. Across all industrial applications, aerobic bacteria are advantageous for their higher growth rates and shorter process times compared to anaerobic. [3] Their use of oxygen as an energy source allows for simple process designs, but also requires optimization for enough oxygen to be supplied without wasted costs. This lab investigated the effect of compressed air flow rate, circulation, and MgSO₄ salt on the rate of enzyme catalase of H₂O₂. Lower flow rates without circulation or salt were found to lead to the most reaction for the cheapest input, but inclusion of the other two variables could be used to optimize certain characteristics of the column depending on the application. High flow rates also had the fastest transfer rates of H₂O₂ to the enzyme, but the benefits of this will have to be compared to the economical costs when applied.

Table of Contents

| List of Symbols | 3 |
|--|----|
| Introduction | 5 |
| Methods | 7 |
| Overview | 7 |
| Treatment of Collected Data | 8 |
| Results and Analysis | 10 |
| Oxygen Concentrations, Mixing Times and Bubble Sizes | 10 |
| Gas Hold Up and Residence Time | 14 |
| Visual Comparison | 17 |
| Discussion and Conclusions | 20 |
| Calculation Examples | 21 |
| Uncertainty Analysis | 22 |
| References | 23 |
| Appendices | 24 |

List of Symbols

| Symbol | Variable | Units | Value (if applicable) |
|------------|---|-----------------|-----------------------|
| K_1a | Volumetric Mass Transfer Coefficient | s ⁻¹ | - |
| OTR | Oxygen Transfer Rate | mmol/(L*s) | - |
| ε | Gas Hold-Up | % | - |
| v_G | Superficial Gas Velocity | m/s | - |
| τ | Gas Residence Time | S | - |
| V_G | Gas Volume | m ³ | - |
| V_{Liq} | Liquid Volume | m ³ | - |
| т | Volumetric Flow Rate | m³/s | - |
| С | Concentration | mmol/L | - |
| Н | Liquid Height in Column | m | - |
| Δp | Pressure Difference | Pa | - |
| D | Column Diameter | m | 0.592 |

| A_C | Column Cross Sectional Area | m² | 0.275 |
|-------|---|----------|-----------------------------|
| g | Gravitational Acceleration | m/s² | 9.8 |
| ρ | Water Density | kg/m³ | 997.05 |
| σ | Water Surface Tension | kg/s² | 0.072 |
| η | Water Dynamic Viscosity | kg/(s*m) | 8.9*10 ⁻⁴ |
| Во | Bond Number | - | $\frac{gD^2\rho}{\sigma}$ |
| Ga | Galilei Number | - | $\frac{gD^3\rho^2}{\eta^2}$ |
| Fr | Froude Number | - | $rac{v_G}{\sqrt{gD}}$ |
| | Pump, pointing in the direction of flow | - | - |
| | Globe Valve | - | - |
| | Manual Gate Valve | - | - |

| | Aeration Nozzle | - | - |
|--------------|-----------------|---|---|
| \ | | | |
| | | | |
| | | | |
| | Oxygen Sensor | - | - |
| (Al1) | | | |
| | | | |
| | | | |

Introduction

Fermentation is the most widely known use of bacteria in industrial processes.^[6] The beverage industry has for centuries used fermenting bacteria to produce alcohol using carbohydrates as an energy source. Treatment and purification of water can also involve several strains of bacteria that breakdown organic molecules in their fermentation cycle.^[6] While yeast is the most important microorganism in the baking industry, several types of bacteria are also used for conditioning and softening of dough via fermentation.^[6]

In the fermentation process, aerobic bacteria in solution require a critical concentration of dissolved oxygen (DO) in order to survive. [2] In the environment, diffusion of oxygen into liquids containing bacteria is generally fast enough to maintain their lifecycle. For industrial applications, aerobic bacteria are used in much higher concentrations in order to take advantage of their fast process times and growth rates. This requires oxygen be actively added to operation steps involving bacteria, so that other factors affecting their reaction rate may then be optimized. In most cases, the second rate limiting step to the bacteria's metabolic reaction is the availability of their food source. [2]

A typical method of increasing DO levels is through bubble aeration, where oxygen is released in the bottom of a column containing fermenting bacteria. As opposed to mixing fans or other exchange equipment, aerators require much less energy to operate and can have other fluids released alongside the oxygen stream. This is often done if the reactant for the bacteria's metabolism is a liquid, resulting in two-fluid nozzle aeration. When two-fluid aeration is incorporated into a process in this way, it can also save the additional step of providing the bacteria with their food source from some other equipment.

In this lab, the enzyme Catazyme, from Novozymes, was used to simulate the metabolism of bacteria. It reacts with hydrogen peroxide to produce water and oxygen, which was measured via the hydrogen peroxide-catalase method. In this method, H_2O_2 was flowed from the two-fluid nozzle and oxygen concentrations were recorded as the enzyme reacted with the chemical. This allowed for the mass transfer coefficient and oxygen transfer rate (OTR) to be found, which are important measurements of how well the H_2O_2 is mixed in the column. Better mixing allows for more of the desired reaction to take place quicker, but should be performed with as little energy and material input as possible. The variables tested affecting this were the flow rate of compressed air, the inclusion of a circulation system underneath the column, and the presence of MgSO₄ salt in the solution.

It was found that low air flow rates without circulation led to the highest amount of reaction from the enzyme. Considering the costs incurred from increasing air flow rate or including the other variables, this should be considered the best general use of the column. High flow rates greatly increased the mass transfer rate, but depending on the specific application this may not be worth investment. The circulation and presence of salt were found to change certain other derived values which may be useful depending on the application. Both circulation and salt were found to lower the mixing time and increase the gas hold up, which can be useful for fast paced processes. The differences between these two methods should be investigated further to determine which is more suited to a specific application. Results should also be repeated with use of live bacteria to confirm the results found in this lab.

Methods

Overview

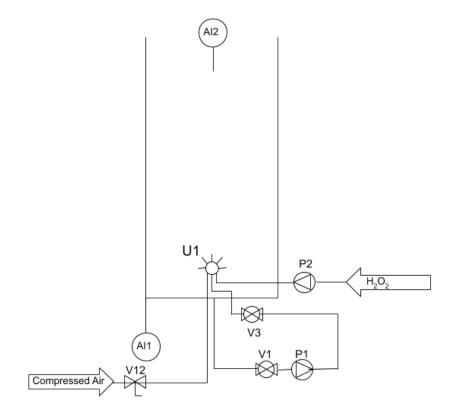


Figure 1. Bubble aeration column used in lab.

Figure 1 shows the water column used in the lab, which could release both compressed air and hydrogen peroxide through aeration nozzle U1. The flow rates of each could be controlled through manual valve V12 and pump P2, respectively. These were varied to determine the effect they had on the mass transfer of oxygen from the liquid to the gas phase, where it was respired by the bacteria. Higher flow rates would increase the amount of dissolved oxygen, but once it was above the critical value needed for respiration then extra pumping was a waste of energy. The unit also had an underchamber with circulation pump P1 that could be connected or shut off from the rest of the column through valves V1 and V3. Circulation uses more energy, but was also investigated for its effect on metabolism and DO levels. Finally, MgSO₄ heptahydrate was added at 0.04 M through the column and circulation pipes.

Table 1. Conditions for each test ran during the lab.

| Test | Compressed Air Flow Rate (L/min) | | |
|------|-------------------------------------|-----|------|
| 1 | 300 | 0 | 0 |
| 2 | 600 | 0 | 0 |
| 3 | 300 | 100 | 0 |
| 4 | 600 | 100 | 0 |
| 5 | 300 | 0 | 0.04 |
| 6 | 300 | 100 | 0.04 |

Table 1 lists the six tests that were performed during the lab. These varied the flow rate of compressed air, the use of the circulation pump, and the presence of the MgSO₄ salt to determine the optimal conditions for operation. Each test began once flow of hydrogen peroxide was introduced, which was constant at 1.9 g/s.

Treatment of Collected Data

Oxygen Concentrations, Mixing Times, and Bubble Sizes

As previously mentioned, DO concentrations must be above a critical value for respiration to take place, but not so high that unnecessary energy costs are incurred from the air pumps. Mixing conditions also affect H_2O_2 transfer to the bacteria and the reaction rate once DO levels are above the critical value. Sensors Al1 and Al2 in **Figure 1** recorded the percent of gas relative to the saturated concentration value. This data was then multiplied by the solubility of oxygen under the conditions of each sensor for each test, which is derived in the **Appendices**. The results show how each test condition affected the rate and magnitude of H_2O_2 transfer and thus the catalase reaction. An estimate of the mixing time was also found from how long each condition took to reach 95% of its steady state concentration value.

$$\frac{d_{vs}}{D} = 26 * Bo^{-0.5} * Ga^{-0.12} * Fr^{-0.12}$$
 Eq. (1)

Eq. 1 was then used to find the average bubble diameter d_{vs} from the column diameter D, the Bond number Bo, the Galilei number Ga, and the Froude number Fr. It is derived from a force balance between coalescence and break up of the bubbles, which reduces its applicability only to coalescing fluids. Because salts act as inhibitors to coalescence at 0.032 M, the concentrations used in this lab prevent **Eq. 1**'s applicability to those trials.

Gas Hold-Up and Residence Time

The main power requirement from bubble aerators comes from pumping the gas at the bottom of the column against the pressure of the liquid on top of it. The fraction of the gas to total volume in the column, known as the gas hold-up ε , is a measure of the intensity of the contact the gas has with the liquid. Lower gas hold-ups show the liquid's high pressure forces the gas to a smaller volume, while the higher values show the opposite. This was measured in the lab by recording the height of the column with and without the aerator flowing for each condition.

$$\varepsilon = 0.6 * v_G^{0.7}$$
 Eq. (2)[8]

Eq. 2 gives an empirical relationship for predicting gas hold up for coalescing air-water systems from the superficial gas velocity v_G . The trials containing MgSO₄ salt are also not applicable to this equation due to their prevention of coalescence.

Gas residence time is defined as the average time it takes for the gas to pass through the liquid column. This is important for understanding mass transfer, as higher residence times can lead to more of the fluid interacting with the enzyme in the column.

$$\tau = \frac{H}{v - c}$$
 Eq. (3)

The gas residence time can be calculated using **Eq. 3**, where H is the height of the liquid and gas mixture in the column and v G is the linear superficial gas velocity.

Transfer Rates

A direct way to quantify the transfer of the H_2O_2 from the pumped fluid to the enzymes is by the volumetric mass transfer coefficient K_1a . This is a value that represents the influence of transfer resistances in the system, so that when resistance to transfer is large the K_1a value is small. In practice, the goal would be to maximize its value so more transfer can take place in the system.

$$K_1 a = \frac{m * c_{peroxide}}{2V(c - c^*)}$$
 Eq. (4)

Experimental K_1a values were determined from **Eq. 4** using the O_2 concentrations found by sensors and the saturated concentration derived in the **Appendices**. The concentration of the hydrogen peroxide used during the lab was found from titration with KMnO₄, while the volumetric flow rate of H_2O_2 m and volume of the reactor V are known.

$$OTR = K_1 a * (c^* - c)$$
 Eq. (5)

The oxygen adsorption/transfer rate (OTR) is given by **Eq. 5**. It is related to the volumetric mass transfer rate with the exception that it does not depend on the oxygen concentrations found during the lab, as multiplying them by the K_1a cancels them out in the denominator of **Eq. 4**. It does result in the OTR being multiplied by -1, which changes the direction of the transfer and implies the oxygen is being respired by the enzyme. The value is important to measure as it shows how much oxygen would be needed for the enzyme to maintain higher levels of the catalase reaction, affecting operation costs.

$$\frac{K_1 a*A}{Q} \left(\frac{\eta^2}{\rho^2 g}\right)^{1/3} = \left(2.4*10^{-6}\right) * \left(\frac{\Delta p*m}{Q\rho \left(\frac{g\eta}{\rho}\right)^{2/3}}\right)^{0.33}$$
 Eq. (6)

Theoretical values for $K_{1}a$ can be calculated from **Eq. 6** for comparison. It uses the physical dimensions of the column along with the properties of the liquid inside. The relationship is also only valid for coalescing liquids.

Results and Analysis

Oxygen Concentrations, Mixing Times and Bubble Sizes

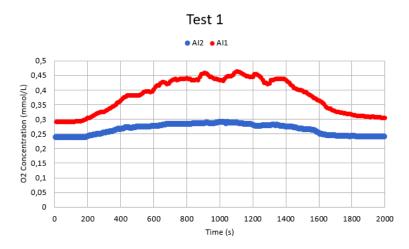


Figure 2. Oxygen concentration over time for low air flow and no circulation or salt.

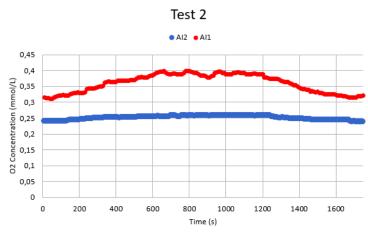


Figure 3. Oxygen concentration over time for high air flow and no circulation or salt.

Changes in oxygen concentration over time for each test indicate how much of the desired reaction with H_2O_2 is taking place. In most industrial settings, direct measurements of reaction materials like this could be the first estimate of whether or not the system is meeting performance expectations. **Figures 2** and **3** show the effect switching from low to high air flow

rate with no influences from circulation or salt. Higher air flow rates showed lower overall concentrations of O₂, indicating less of the hydrogen peroxide was reacted.

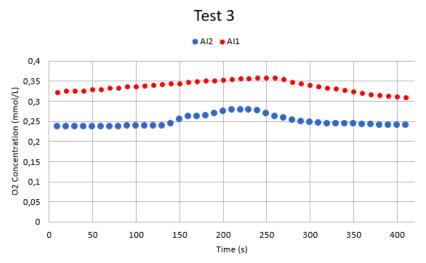


Figure 4. Oxygen concentration over time for low airflow, circulation, and no salt.

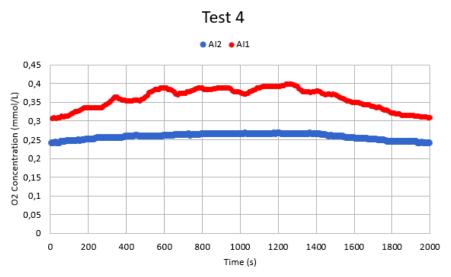


Figure 5. Oxygen concentration over time for high air flow, circulation, and no salt.

Figures 4 and 5 repeated the tests of Figures 2 and 3 but with circulation included. In this case, the circulation had the effect of lowering the oxygen concentrations in the low air flow rate test while the high air flow rate was mostly unaffected. Since the H_2O_2 was mixed with the air before release through the nozzle, the higher flow rate may have allowed more of it to move up the column because of the higher velocity at which it escaped. H_2O_2 in the lower air flow rate may have been more affected by the circulation at the bottom, leading it to not be as well dispersed throughout the column. Since the circulation is meant to increase the mixing while using energy to do so, a lower amount of reaction would then easily dismiss it from consideration.

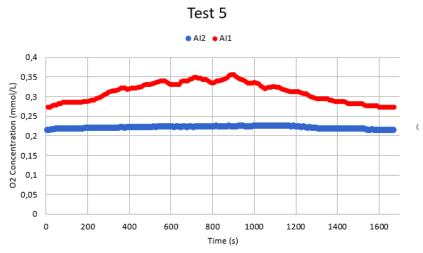


Figure 6. Oxygen circulation over time for low air flow, no circulation, and salt.

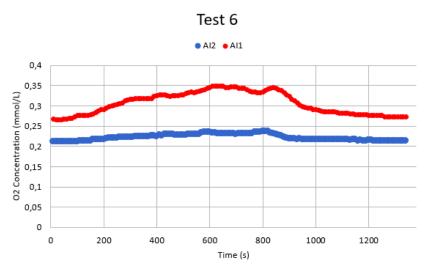


Figure 7. Oxygen circulation over time for low air flow, circulation, and salt.

Figures 6 and **7** show the effect of circulation when low air flow and salt was used in the column. In the two, the oxygen concentrations were mostly the same although the circulated test had a slightly lower maximum. Both of these tests, like **Figure 4**, showed overall much lower oxygen concentrations than **Figure 2**, where low air flow was used without circulation or salt. Since **Figure 2** also demonstrates the cheapest case, where extra power and material won't have to be supplied to the system, the higher amount of produced oxygen makes it considerable for the best alternative.

Table 2. Mixing times for each test condition.

| | Low Air Flow Rate | | | | High Air f | low Rate |
|-----------------|----------------------------|---------|-------------------------|---------|------------|-------------|
| | Circulation No Circulation | | ation No Circulation | | | Circulation |
| | No Salt | Salt | | No Salt | | |
| Mixing Time (s) | 145 <u>±</u> 40 | 255±300 | 255±300 320±500 725±200 | | 380±200 | 385±200 |

Table 2 demonstrates the changes in the mixing time for each set of conditions tested. The conditions of **Figure 2** had the longest mixing time, while the shortest, **Figure 4**, differed from the former only by the addition of circulation. The addition of the salt also had mixed effects on the mixing time. With low air flow, it raised the value when there was circulation while without circulation it lowered it.

Previously it was considered that **Figure 1** may offer the largest amount of reaction for the cheapest cost, but if less time is spent getting to steady state reaction conditions then the addition of circulation would make the operation more time effective. The application of the bubble aerator also would affect which type of performance is chosen. If only a smaller amount of product is needed from the enzyme, or the unit is part of a fast paced process, then the addition of circulation would fulfill these objectives in a shorter time. If the unit is in a process with a more expensive reactant, or the reaction needs to proceed more fully to completion, then excluding the circulation would be more applicable.

Table 3. Average bubble diameters for test conditions with no salts from Eq. 1.

| | Low Air F | low Rate | High Air Flow Rate | | |
|---------------------------------|----------------|-------------|--------------------|----------------|--|
| | No Circulation | Circulation | | No Circulation | |
| Average Bubble Diameter (mm) | 4.1±0 | 4.1±0 | 3.8±0 | 3.8±0 | |

Table 3 gives the average bubble sizes for the tests that were applicable from **Eq. 1**. Bubble size was dependent only on the superficial velocity, and thus flow rate of the air. Higher flow rates led to smaller bubbles while lower flow rates allowed larger bubble growth as the air left the nozzle. Larger bubbles in general lead to coalescence, which tends to lower mass transfer. If mass transfer is desired to be as large as possible, higher flow rates should be used to promote this. In order to optimize this variable, a more detailed study could also determine the cost trade off of increasing the flow rate while increasing mass transfer and find the point where it is best fit for the operation.

Table 4. Experimental and theoretical gas hold up values, as a percentage, for each set of conditions tested. Theoretical calculations come from **Eq. 2**.

| | Low Air Flow Rate | | | | High Air f | low Rate |
|--------------|----------------------------|----------------|---------------------|------------------|------------------|------------------|
| | Circulation No Circulation | | tion No Circulation | | | Circulation |
| | No Salt | Salt | | No Salt | | |
| Experimental | 7.0 <u>+</u> 1 | 9.2 <u>+</u> 1 | 5.2 <u>+</u> 1 | 5.2 <u>+</u> 1 | 13 <u>+</u> 1 | 15 <u>+</u> 1 |
| Theoretical | 3.6 <u>±</u> 0.0 | - | - | 3.6 <u>±</u> 0.0 | 5.9 <u>+</u> 0.0 | 5.9 <u>±</u> 0.0 |

Table 4 shows the gas hold up values found from experiment and from **Eq. 2**. Larger gas hold ups are advantageous as it means the gas had a higher volume and contact with the liquid. From the experiment, higher air flow rates led to the largest gas hold up values. Among both flow rates, adding circulation increased the value. The addition of salt also led to a larger gas hold up when circulation was used, but did not affect the case without circulation. Non coalescing liquids, such as in the salt trials, may be estimated to have hold ups of up to 50-200 times as large compared to coalescing liquids in general. This would suggest the salt trials would perform better, but since this is only found in one of the experimental tests it is inconclusive. Since **Eq. 2** was only affected by the flow rate, higher flow rates were estimated to have a larger gas hold up, which agrees with the experimental results.

While higher gas hold ups are desirable in general, it is not on its own what bubble aerators should be designed for. Both the oxygen concentrations for each test and the mixing times previously mentioned should be considered more important results as they more directly relate to the enzyme reaction. Gas hold up may be used as a general indicator for contact between the gas and liquid in the column, but the other derived values from the experiment more meaningfully quantify this. The highest gas hold up conditions still generally correspond to the optimal conditions for mixing time and bubble size, as higher flow rates and circulation generally improved performance.

Table 5. Gas residence times for each test condition, calculated using **Eq. 3**.

| | Low Air Flow Rate | | | | High Air F | Flow Rate |
|------------------------------|-------------------|----------------------|----------------|----------------|---------------|---------------|
| | Circul | ation No Circulation | | | n | Circulation |
| | No Salt | Salt | | No Salt | | |
| Gas Residence Time (s) | 119 <u>+</u> 3 | 123 <u>±</u> 3 | 116 <u>+</u> 3 | 116 <u>+</u> 3 | 63 <u>±</u> 1 | 66 <u>±</u> 1 |

The gas residence time for each test condition can be seen in **Table 5**. These results show that the most important factor in gas residence time is the air flow rate. In the tests that were conducted using a high flow rate, the gas residence time is about half as long as it is for the tests performed with a low flow rate. As the air flow rate is increased, the gas residence time decreases because the gas is flowing faster, and therefore passes through the column quicker. It can also be seen that salt has very little, if any, effect on gas residence time. Two tests, one with salt and one without, with all other conditions being the same, demonstrated that the gas residence time was equivalent. This means that the addition of salt has no influence on the time it takes the gas to pass through the column. Lower residence times may be preferred in industry if longer contact is desired between the enzymes and aerated fluid, or shorter times may be preferred if the reaction can proceed fast enough that it is not the rate limiting step. If a reaction is desired to proceed at a specific rate, and the residence time of the gas is rate limiting, then the air flow rate may be varied to achieve the target speed that would lead to the specific time.

Transfer Rates

Table 6. Theoretical and experimental K_1a and experimental OTR values for each test condition, calculated using **Eqs. 4, 5, and 6**.

| | Low Air Flow Rate | | | | High Air Flow Rate | |
|---|-------------------|------------------|----------------------|------------------|--------------------|------------------|
| | Circul | ation | ation No Circulation | | | Circulation |
| | No Salt | Salt | | No Salt | | |
| Theoretical K 1a Value (s-1) | 0.015 <u>+</u> 0 | ı | ı | 0.015 <u>+</u> 0 | 0.023 <u>±</u> 0 | 0.023±0 |
| Experimental K 1 a Value (s ⁻¹) | 0.14 <u>±</u> 0 | 0.076 <u>±</u> 0 | 0.082 <u>+</u> 0 | 0.043 <u>+</u> 0 | 0.073 <u>±</u> 0 | 0.062 <u>±</u> 0 |
| OTR (mmol/(L*s)) | 5.4 <u>±</u> 0 | 5.3±0 | 6.2 <u>±</u> 0 | 6.2 <u>±</u> 0 | 5.7 <u>±</u> 0 | 4.9±0 |

Table 6 gives the experimental and theoretical volumetric mass transfer rate along with the OTR for each set of conditions. The K_1a values indicate more transfer per unit time when they are larger, with OTR proportional to K_1a by **Eq. 5**. In general, experimental values were higher than the theoretical, although this was probably due to differences in the aeration column from where **Eq. 6** was derived. For example, interactions between the two fluids being aerated are not included by the equation, and they may have an affect on the transfer into the water. The same general trends are observed, however, such as the higher flow rate leading to larger amounts of transfer per time. While the effect of adding salt could not be modeled by the correlation, in the experiment it also greatly increased the mass transfer, making it more influential than the high flow rate. Circulation slightly reduced mass transfer with a high flow rate, while it greatly reduced the transfer for the low flow rate. The OTR, as a result, followed the same trends as the K_1a values.

Without other considerations, processes should generally try to increase the rate of mass transfer as it saves time during the process. Time savings are also financial savings when the operation requires continuous pumping, as in all cases of bubble aeration. The best transfer would be judged to be the low flow rate with salt based on these results, as it requires less pumping power while also achieving faster mass transfer than the high air flow rate. The differences between the OTR values were less great than the K_1a , but would also agree with this result. If a process could not use a salt that increased the transfer rate, such as from sensitivity in the bacteria or enzyme, then higher air flows would need to be considered for their higher performance with additional cost.



Figure 8. Photos of low air flow rate, left, and high air flow rate, right, both with no circulation and salt.

Figure 8 shows how an increase in air flow rate lead to much more bubbles being produced than without. This agrees with the results of **Table 3**, which predicted smaller bubble sizes, and thus more bubbles, from the high air flow rate. Since transfer is occuring from the interface of each of these bubble's to the surrounding liquid, it can be seen how higher flow rates led to faster transfer as in **Table 6**.



Figure 9. Photos of low air flow rate, left, and high flow rate, right, with circulation and no salt.

Figure 9 repeats the conditions of **Figure 8** but with circulation. Generally, the amount of cloudiness from bubbles around the base of the column is reduced. From **Table 2**, this change was seen to decrease the mixing time compared to the two previous photos. This most likely comes from the larger amount of contact the stream has with the water near the nozzle, as it would collide less with other static bubbles near the bottom. The extra air that is still present in the high air flow condition also supports the larger gas hold ups that were found for those cases in **Table 4**. The more bubbles that are seen on the right would lead to a larger volume of gas in the column, and thus higher measured column height.



Figure 10. Photos of low air flow rate with no circulation, left, and circulation, right, and salt.

Figure 10 demonstrates the effect of salt when added to the low air flow rate condition with and without circulation. The combined effect of circulation and salt are seen to drastically improve the visibility through the lower part of the column, which also gave one of the quickest mixing times tested. If it was critical that the fluid being included in the aeration column be encountered quickly by a bacteria or enzyme, this condition would be the most preferential. Such a case could happen if the fluid degraded with air on the same time scale as the mixing time, or if there was a step immediately before or after this process that was sensitive to time.

Discussion and Conclusions

When choosing the best conditions to run the bubble aerator, the largest consideration should be for what advantages each variable provides the operation for their costs. Different aspects of the column, such as the total fluid mixed, the speed at which mixing occurs, or the efficiency of the mixing, could be prioritized differently based on the application. The lowest cost option, that with low air flow rate and no circulation or salt, was found to lead to the greatest amount of catalase reaction with the H_2O_2 , as shown by the largest O_2 concentrations in **Figure 2**. The two most expensive conditions tested, that being high flow rate or low flow rate with circulation and salt, led to the highest mass transfer rates as well as fastest mixing times. By completing the mixing faster, the unit may need to be operated for less time, thus making the unit more economical as well as time saving.

Still, the high flow rate option still could not reduce the mixing time by greater than a factor of two when compared to the longest low flow rate test. That is important in this experiment as the high flow rate was pumped at twice the flow rate of the low, thus requiring twice the power. To make it economical in this context, the unit needed to cut down the required time by at least 50%, which it did not. If there were additional reasons for a specific application that a shorter time would be preferable, it may make it worthwhile, but in this context it was not. Large uncertainties were present for these values, however, which may warrant repeating these tests for comparison.

The largest contributor to the uncertainty for these trials was from the different readings the oxygen sensors gave. In all cases of measured oxygen concentrations, the Al1 sensor reported much higher oxygen concentrations than Al2, as the former was submerged at the base while the latter was exposed to atmosphere ~3 m high in the column. It can be seen that Al2 was less affected than Al1 through **Figures 2-7**, and stayed more consistently around the atmospheric concentration of oxygen in air at 0.21 mmol/L. If a column were to be sealed as part of a process, or completely filled with liquid, these results should also be reevaluated to determine how values may differ from the bottom to the top of the column. For industrial applications, columns may be twice the size of those used in the pilot plant and could have considerable spatial differences from the nozzle to the top.

Other values and correlations for K_1a from literature were not found applicable to what was performed in lab, but could also be evaluated if the design of the experiment was changed. For example, Akita and Yoshida report a correlation for determining K_1a from the gas hold up and time required for complete liquid mixing in the column. Since H_2O_2 was continuously pumped during this experiment, the complete mixing time was not determined. From the same source, the gas hold up for water-air systems was also reported to be in the range of 0.038-0.171 in the range of 20-30C. This is covers all experimental values which were found in this lab. In addition to the theoretical correlations already used for analysis of this lab, the agreement with other sources is a good sign that results can be used for further evaluation or scale up of this unit.

Calculation Examples

All example calculations use data from Test 1.

Bubble Diameters

Eq. 1 gives the average bubble diameter based on an empirical relation between the diameter of the column and three dimensionless groups that characterize the flow conditions.

$$d_{vs} = D * 26 * Bo^{-0.5} * Ga^{-0.12} * Fr^{-0.12}$$
 Eq. (1)

$$d_{vs} = 0.592 \ m * 26 * 47,500^{-0.5} * 2.56E12^{-0.12} * 0.0075^{-0.12} = 4.1 \ mm \quad \text{Eq.} \eqno(1)$$

Gas Hold Up and Residence Time

$$v_G = \frac{m}{A_C} = \frac{0.00500 \, m^3/s}{0.275 \, m^2} = 0.0182 \, m/s$$
 Eq. (7)

Eq. 7 gives the superficial linear velocity of the gas from the pumped volumetric flow rate and the cross sectional area of the column.

$$\varepsilon = 0.6 * v_G^{0.7} = 0.6 * 0.0182$$
 $^{0.7} = 0.036$ Eq. (2)

Eq. 2 gives the theoretical gas hold up directly from v_G .

$$\varepsilon = \frac{V_G}{V_G + V_{Liq}} = 5.2$$
 Eq. (8)

Eq. 8 shows the calculation of experimental gas hold up from the volumes of gas and liquid in the column. These were found by measuring the height difference in the column before and after aeration for each test.

$$\tau = \frac{H}{v_{G}} = \frac{2.11 \, m}{0.0182 \, m/s} = 116 \, s$$
 Eq. (3)

Eq. 3 finally gives the residence time from the height of the column and v_G .

Transfer Rates

$$\frac{K_1 a*A}{Q} \left(\frac{\eta^2}{\rho^2 g}\right)^{1/3} = (2.4*10^{-6}) * \left(\frac{\Delta p*m}{Q \rho \left(\frac{g\eta}{Q}\right)^{2/3}}\right)^{0.33}$$
 Eq. (6)

Eq. 6 gives the theoretical values for $K_{1}a$ using the physical dimensions of the column as well as the properties of the liquid inside.

$$\frac{K_1 a * 0.275}{0.01818} \left(\frac{(8.9 \cdot 10^{-4})^2}{997.05^2 g}\right)^{1/3} = \left(2.4 * 10^{-6}\right) * \left(\frac{40000 * 1.31 \cdot 10^{-6}}{0.01818 * 997.05 \left(\frac{9.8 * 8.9 \cdot 10^{-4}}{997.05}\right)^{2/3}}\right)^{0.33}$$
Eq. (6)

$$K_1 a = 0.015$$
 Eq. (6)

Eq. 6 gives the theoretical value for K_1a in the bubble column process.

Uncertainty Analysis

All examples use values from Test 1 when applicable.

Tests for each set of conditions were only performed once, so uncertainty between multiple samples is not present. The exception to this was for determining the mixing time of each condition. Uncertainty for this value comes from the two sensors that were used during the experiment. As they each made independent measures of concentration over time, two different values of mixing time were found for each run. The standard deviation of these two values was then presented as the uncertainty, along with an estimated one percent error in the sensors ability to detect the oxygen concentrations. The estimate of the one percent error came from their random variation when no changes were being made to the column.

Uncertainty that the rest of the equipment displayed correct values was, however, present for all analysis. The flow rate of the compressed air could only be measured to ± 5 L/min, which results in an uncertainty of 0.3 mm/s for measurements of superficial velocity. This assumes that column dimensions were known exactly.

$$\sigma_{\varepsilon} = \sqrt{\sigma_{v_G}^2 * (\frac{d\varepsilon}{dv_G})^2} = \sqrt{\sigma_{v_G}^2 * (0.42 * v_G^{-0.3})^2}$$
 Eq. (9)

$$\sigma_{\varepsilon} = \sqrt{(0.0003)^2 * (0.42 * 0.0182^{-0.3})^2} = 0.042\%$$
 Eq. (9)

Eq. 9 gives the uncertainty for theoretical gas hold up values based on the uncertainty of v_G . This comes from the nonlinear relationship between the two shown in **Eq. 2**. Since the uncertainty is smaller than the last significant figure in the value, the uncertainty is not significant. The bubble diameter and K_1a/OTR calculations were also functions of only one uncertain variable, which was found via the same method as **Eq. 9**. The former had uncertainty from v_G while the latter had it from the repeated titrations.

Uncertainty in the measurement of the height of the column also introduces uncertainty in the experimental gas hold up.

$$\sigma_{\varepsilon} = \sqrt{\sigma_{V_G}^2 * \left(\frac{d\varepsilon}{dV_G}\right)^2} = \sqrt{\sigma_{V_G}^2 * \left(\frac{V_{Liq}}{\left(V_{Liq} + V_G\right)^2}\right)^2}$$
 Eq. (10)

$$\sigma_{\varepsilon} = \sqrt{(0.0137 \ m^3)^2 * (\frac{0.551 \ m^3}{(0.551 \ m^3 + 0.302 \ m^3)^2})^2} = 1.3$$
 Eq. (10)

Eq. 10 shows this uncertainty based on the volume of the gas V_G , which was what uncertainty in the height of the column affected. Recorded heights of the fluid level in the column were estimated to have an uncertainty of ± 0.03 m. This is because the tape measure on the column was read from a considerable distance when standing on the plant floor.

$$\sigma_{\tau} = \sqrt{\sigma_{H}^{2} * \left(\frac{d\tau}{dH}\right)^{2} + \sigma_{v_{G}}^{2} * \left(\frac{d\tau}{dv_{G}}\right)^{2}} = \sqrt{\sigma_{H}^{2} * \left(\frac{1}{v_{G}}\right)^{2} + \sigma_{v_{G}}^{2} * \left(\frac{-H}{v_{G}^{2}}\right)^{2}}$$
 Eq. (11)

$$\sigma_{\tau} = \sqrt{(0.03 \ m)^2 * (\frac{1}{0.0182 \ m/s})^2 + (0.0003 \ m/s)^2 * (\frac{-2.11 \ m}{(0.0182 \ m/s)^2})} = 2.5 \quad \text{Eq. (11)}$$

Finally, **Eq. 11** shows combined uncertainty in the residence time as a function of both uncertainty in the height of the column as well as in the air flow rate.

References

- [1] Andreas L and Murillo VF. Production of fine chemicals using biocatalysis. *Current Opinion in Biotechnology*. 1999;10(6):595-603. doi:10.1016/S0958-1669(99)00040-3.
- [2] DTU Chemical Engineering. Large Scale Exercises in Process Technology and Chemical Unit Operations Summer 2019.
- [3] Hatti-Kaul, Rajni and Mattiasson, Bo. Anaerobes in Industrial- and Environmental Biotechnology.

Anaerobes in Biotechnology. 2016:1-33. doi:10.1007/10 2016 10

- [4] Jozala AF, Geraldes DC, Tundisi LL, et al. Biopharmaceuticals from microorganisms: from production to purification. *Braz J Microbiol*. 2016;47 Suppl 1(Suppl 1):51–63. doi:10.1016/j.bjm.2016.10.007
- [5] Najafpour, Ghasem. (2007). Industrial Microbiology. 10.1016/B978-044452845-2/50001-X.
- [6] Singh R, Kumar M, Mittal A, Mehta PK. Microbial enzymes: industrial progress in 21st century. *3 Biotech*. 2016;6(2):174. doi:10.1007/s13205-016-0485-8
- [7] Stanbury, P.F. et al.: Principles of Fermentation Technology. Butterworth Heinemann, 2. Ed.
- [8] Deckwer, W.-D.: Bubble Column Reactors. Wiley, 1992.
- [9] Akita, Kiyomi and Yoshida, Fumitake. *Industrial & Engineering Chemistry Process Design and Development* 1973 *12* (1), 76-80. doi: 10.1021/i260045a015

Appendices

Oxygen Concentrations

Saturated O_2 solubilities were found from literature to be 1.26 mmoles/L in pure water and 1.12 mmol/L in water with 2% NaCl, both at 25C and 1 atm of O_2 pressure.^[7] The latter solubility was used as an estimate for the effect of adding MgSO₄ salt to the mixture.

| Table 7. Saturated concentrations | s of O_2 in water, with | n and without aqued | ous salt, for the | | |
|---|---------------------------|---------------------|-------------------|--|--|
| pressures of the two sensors. Data is presented at 25 C.[7] | | | | | |
| | | | | | |

| Sensor | Pressure (atm) | O₂ Partial Pressure (atm) | Saturated O ₂ Concentration in Pure Water (mmol/L) | Saturated O ₂ Concentration in Water + Salt (mmol/L) |
|--------|----------------|------------------------------|---|---|
| Al2 | 1 | 0.21 | 0.27 | 0.24 |
| Al1 | 1.2 | 0.25 | 0.32 | 0.28 |

Table 7 shows the final saturated concentrations of oxygen that were multiplied by the sensor readings to generate the solubility over time curves. Because Al1 was under ~2 meters of water, it was under an increase of pressure of about 0.2 atm compared to Al2. This resulted in slightly higher saturated solubility concentration at the bottom of the column than at the liquid surface.

Extended Uncertainty Derivations

Uncertainty in the gas velocity introduced uncertainty in the calculation of mean bubble diameter.

$$\sigma_{dvs} = \sqrt{\sigma_{v_G}^2 * (\frac{ddvs}{dv_G})^2}$$
 Eq. (12)

$$= \sqrt{\sigma_{v_G}^2 * (D * 0.26 * Bo^{-0.5} * Ga^{-0.12} * \frac{-0.12 * v_G^{-1.12}}{\sqrt{gD}})^2}$$
 Eq. (12)

$$\sqrt{(0.003 \, m/s)^2 * (0.592 \, m * 0.26 * 47,560^{-0.5} * 2.55E12^{-0.12} * \frac{-0.12*0.0182m/s^{-1.12}}{\sqrt{9.8m/s^2*0.592m}})^2}$$

$$= 30 \, nm$$
Eq. (12)

This uncertainty in **Eq. 12** is insignificant compared to what is presented in **Table 3**, being several orders of magnitude smaller than the bubble diameters on the scale of millimeters.